time to relapse from day 30 post-transplant was 69 days (range 41-230 days). The cumulative incidence of acute GVHD grade II-IV was 44% with 10% grade III-IV. The cumulative incidence of chronic GVHD at 2 years was 48%, with 22% extensive chronic GVHD.

Conclusion: In this cohort of patients with AML/ MDS, MCP-1 levels at day 30 post allogeneic HCT in patients who had achieved complete remission and full chimerism were predictive of relapse two months on average prior to overt hematological relapse. Larger studies may find potential role of MCP-1 in predicting relapse post-transplant and development of early strategies for prevention.

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Targeting BCL-2 and BCR-Abl Activity in Ph+ALL

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Background: Treatment of adult Philadelphia chromosome positive acute lymphoblastic leukemia (Ph^+ALL) remains a challenge. Tyrosine kinase inhibitors (TKI's) have greatly improved treatment options for Ph^+ALL, leading to an increased number of patients eligible for curative hematopoietic stem cell transplant. A current induction regimen for Ph^+ALL is the combination of the targeted dual Abl/Src TKI dasatinib and a corticosteroid. It is thought that corticosteroids work synergistically with TKI's by modulating the BCL-2 family of proteins, leading to apoptosis. However corticosteroids have to be tapered after the first 21 days of induction due to significant toxicities, particularly in older adults. Meanwhile, patients remain on TKI monotherapy until the time of transplant, rendering them susceptible to the development of resistance. Recently BCL-2 inhibitors have been shown to have single agent efficacy in B cell malignancies with relatively low toxicity and our lab has shown *in vitro* effects in primary patient Ph^+ALL samples (unpublished data). Therefore we investigated the potential combination of dasatinib with the BCL-2 inhibitor ABT737 as a targeted combination in Ph+ALL.

Methods: Drug efficacy *in vitro* was determined using the Ph^+ALL cell line SupB15 and the CML cell line K562. Cells were incubated with varying concentrations of dasatinib, ABT737 or in combination for 72 hours. Cell viability was assessed with the colorimetric MTS assay, and synergy was calculated using Calculsyn software. Xenografted cells from a patient with Ph^+ALL were assessed under identical conditions. Apoptosis was assessed with annexin V staining. Expression of the BCL family proteins BCL-2 and MCL-1 were assessed via immunoblot.

Results: The IC_{50} of dasatinib and ABT737 in SupB15 were 8.8nM and 5.9nM, respectively. The IC50 of equimolar combination was 0.42nM, and synergistic with combination index (CI) values between 0.15 and 0.49 (<1=synergy). Primary Ph^+ALL xenograft cells showed a similar pattern of synergy to the dasatinib + ABT737 combination, with CI values between 0.01 and 0.38. Combination treatment increased apoptosis as measured by Annexin V staining. In contrast, the CML cell line K562 was not sensitive to ABT737 as a single agent and there was no enhanced efficacy by adding this agent to dasatinib, suggesting this combination is specific for Ph+ALL. This was supported by increased BCL-2 and low MCL-1 protein expression in SupB15 and xenografted Ph+ ALL cells, whereas K562 had low expression of BCL-2 and high levels of MCL-1.

Conclusions: These data verify that the combination of BCL-2 and BCR-Abl targeting in Ph+ ALL is synergistic *in vitro* laying the foundation for further evaluation *in vivo* for adult Ph^+ALL. Combination targeted therapies may offer the potential for greater and longer responses without the morbidity associated with cytotoxic chemotherapy, particularly in older adults.

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Distinctions Between Effective and Ineffective AML-Specific Autologous Peripheral Blood (PB) Cytotoxic T-Lymphocytes (CTLs)

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Background: Ex-vivo expansion of CBT-cells with CD3/28 co-stimulatory beads, IL-2 & IL-7 & subsequent priming against leukemia cell lines using IL-15 created specific CTLs. [1, 2]

Hypothesis: We hypothesized (a) patient-derived AML-specific PB auto CTLs could be generated (b) T_{regs} proportion ($CD4^+CD25^{bright}FoxP3^+$) & T-cell co-signaling markers' gene expression will be different between effective and ineffective CTLs.

Methods: AML & auto T-cells were purified from PBMC of AML patients admitted with acute blast crisis (n=8). AML blasts were sustained in Serum-Free media (STEMCELL Tech) with MSC support & cytokines (IL-3, SCF, FLT3L, GMCSF, IL-4). T-cells were expanded in culture for 2 weeks as reported [1, 2] & subsequently primed with γ-irradiated auto AML weekly X 3 with IL15 & CD28ab [BD Biosciences]. At the end of week 3 (EOW3), cytotoxicity was assessed against AML and irrelevant targets - IM9 & U937 cell lines, at an E:T ratio of 40:1, 20:1, 10:1 & 5:1 using DELFIA® EuTDA assay.[2] IFN-γ ELISPOT assay against same targets was done.[2] RT-qPCR analysis was done on T-cells before & after priming, with *Power* SYBR® Green master mix (*Thermo Fisher*) & StepOne Plus system (Life Tech). Student *t-test* compared the groups.

Results:

1. T-cells expanded in all samples (n=8) by median of 155-fold (range 11-489) at EOW3.
2. ELISPOT assay was positive in 4/8 samples. [**Fig 1**]
3. CTL assay was difficult to standardize for AML blasts due to high degree of spontaneous apoptosis (>30% spontaneous release [SR]).
   i. 2/8 samples were evaluable (SR<30%).
   ii. Both samples showed AML-specific lysis. [**Fig 2**]
4. Overall, AML-specific auto CTLs could be generated in 5 of 8 samples based on ELISPOT & CTL assays.